scaffold material. A regenerated silk fibroin is first prepared converted to a porous fibroin scaffold using either salt leaching or gas foaming followed in both cases by treatment with methanol or propanol to stiffen and strengthen the material. The material is intended for use as a scaffold for growing a cartilage-like material in vitro. The protocol used for the preparation of the fibroin solution described in US 2007/0187862 involves degumming by boiling cocoons for 20 minutes in an aqueous solution of 0.02 M sodium carbonate solution followed by dissolution of the fibroin in 9.3 M lithium bromide at 60° C. Thus the protocol they used is closely similar to the standard protocol described in the literature and to that used by Holland, C., Terry, A. E., Porter, D. & Vollrath. F. in their paper "Natural and unnatural silks", Polymer 48, 3388-3392 (2007). The latter authors prepared a regenerated fibroin solution prepared according to the standard protocol and compared the rheology of this and native *Bombyx mori* silk fibroin solution taken directly from the silk gland of the silkworm and at the same protein concentration. They concluded that the vast reduction of viscosities and storage modulus values they observed in the regenerated silk fibroin could be explained by degradation of both molecular weight and folding of the fibroin as a consequence of the protocol used. Thus there is strong evidence that the conditions for preparing the fibroin solution disclosed in US 2007/0187862 produce a marked degradation of the fibroin. This is likely to have a markedly negative impact on the compressive strength, moduli and resilience of the porous material produced from the silk fibroin solution.

[0035] WO 2007/020449 teaches an implantable cartilage repair device comprised of a three dimensional biomimetic fibrelay and a bioresorbable porous hydrogel. The hydrogel can be at least partially comprised of regenerated fibroin. The fibroin for the preparation of the porous hydrogel is prepared using the standard protocol comprising the steps of dissolving degummed silk in hot 9.3M lithium bromide solution; dialyzing resulting solution exhaustively against deionised water for two days and concentrating it in a vacuum dessicator.

[0036] US 2005/0281859 describes a method of forming an object from a feedstock, such as fibroin, capable of undergoing a sol-gel transition by adjusting the conditions to cause the feedstock to flow and then adjusting the conditions to gel the feedstock.

[0037] It has recently been shown that porous fibroin hydrogels prepared from the standard protocol are weak and have reduced resilience. Thus, there is still scope for improvement in the implantable materials and implants used for the replacement, partial replacement, or augmentation, or repair of damaged cartilage.

[0038] It is therefore, an object of the present invention to provide an improved regenerated fibroin solution and method of preparing an improved regenerated fibroin solution.

[0039] Another object of the invention is to provide an implantable fibroin material and a method of preparing the fibroin material, having improved mechanical properties.

[0040] It is a further object of the invention to provide an implant for the total or partial replacement, augmentation or repair of cartilage.

SUMMARY OF THE INVENTION

[0041] According to a first aspect of the invention, there is provided a method of preparing a regenerated fibroin solution, the method comprising steps of:

[0042] treating silk or silk cocoons with an ionic reagent comprising an aqueous solution of monovalent cations and monovalent anions, the cations and anions having ionic radii of at least 1.05 Angstroms and a Jones-Dole B coefficient of between −0.001 and −0.05 at 25° C.; and

[0043] subsequently degumming the treated silk or silk cocoons; or alternatively

[0044] degumming silk or silk cocoons; and

[0045] subsequently treating the degummed silk or silk cocoons with an ionic reagent comprising an aqueous solution of monovalent cations and monovalent anions, the cations and anions having ionic radii of at least 1.05 Angstroms and a Jones-Dole B coefficient of between −0.001 and −0.05 at 25° C.

[0046] As will be readily understood by those skilled in the art, the B coefficient of the Jones-Dole equation (Jones, G., and Dole, M., *J. Am. Chem. Soc.,* 1929, 51, 2950) is related to the interaction between ions and water and is interpreted as a measure of the structure forming and structure-breaking capacity of an electrolyte in solution.

[0047] Preferably, the cations and anions have a Jones-Dole B coefficient of between −0.001 and −0.046 at 25° C.

[0048] More preferably, the cations and anions have a Jones-Dole B coefficient of between −0.001 and −0.007 at 25° C.

[0049] It is particularly preferred that the method comprises a further step of drying the silk or silk cocoons after treatment of the silk or silk cocoons with the ionic reagent. Preferably, the drying step is performed consecutively after the step of treatment with the ionic reagent.

[0050] The aim of the drying step is to extract as much water as possible from the treated silk or silk cocoons. Ideally, substantially all of the water is removed from the treated silk or silk cocoons

[0051] The process of drying the silk or silk cocoons may be performed by any suitable means, such as, for example, air drying, freeze drying, or drying through the application of heat.

[0052] Preferably, the step of drying the silk or silk cocoons comprises air drying.

[0053] The silk or silk cocoons may be dried at any suitable temperature. For instance, good results have been observed by drying the silk or silk cocoons at room temperature (21° C.).

[0054] The silk or silk cocoons may be dried over any suitable time period. Typically, the silk or silk cocoons may be dried for a period of several hours, for example 12-16 hours.

[0055] In some embodiments, the silk or silk cocoons may be air dried in conditions of less than 20% humidity. Preferably, drying of the silk or silk cocoons is carried out in the presence of a desiccant, which may include anhydrous calcium chloride or other suitable desiccant. Other suitable desiccants may include silica gel, calcium sulfate, calcium chloride, and montmorillonite clay. Molecular sieves may also be used as desiccants.

[0056] The ionic reagent may comprise a hydroxide solution. The hydroxide solution may be formed in situ. For example, the silk or silk cocoons may be treated with ammonia gas or vapour to form ammonium hydroxide in combination with water already present in the silk or silk cocoons. Further, water vapour may be added to the silk or silk cocoons either before the ammonia gas or vapour, with the ammonia gas or vapour or subsequently.